

Bugs Matter citizen science survey of insect abundance in Gwent

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Abstract

In recent years, scientists and the media have drawn attention to global declines in insect abundance or the so-called 'insect apocalypse', the consequences of which are potentially catastrophic. Invertebrates are critical to ecosystem functions and services, and without them life on earth would collapse. However, there is currently insufficient data to make robust conclusions about trends in insect abundance in the UK, because standardized insect sampling approaches are not widely applied to all insect groups or at a national scale. Here, we demonstrate the use of an innovative and scalable invertebrate sampling technique conducted by citizen scientists, to examine the difference in invertebrate abundance in Gwent over a 17 year timeframe. The 'windscreen phenomenon' is a term given to the anecdotal observation that people tend to find fewer insects squashed on the windscreens of their cars now, compared to in the past. This observation has been ascribed to major declines in insect abundance. In this study, citizen scientists were asked to record the numbers of squashed insects and other invertebrates on their vehicle number plates following a journey, having first removed any residual insects sampled on previous journeys. We compared the number of insects sampled by vehicles in Gwent in 2021 (n = 175 journeys) with the Gwent results of a national survey using this methodology led by the RSPB in 2004 (n = 112 journeys). The results show that the number of insects sampled on vehicle number plates in Gwent decreased by 39.8% between 2004 and 2021, and this difference was statistically significant. This is consistent with the declining trends in insect abundance widely reported by others, and informs a growing requirement for conservation research, policy and practice targeted at invertebrates in the UK. However, our results are based on data with low temporal resolution and consequently we interpret this short-term pattern with caution. Furthermore, inter-annual variation in a range of unmeasured factors, such as wind speed, predation or land-use change, could significantly influence the observed pattern. To draw robust conclusions about long-term trends in insect abundance in Gwent, scientists require data from multiple years, over long time periods, and over large spatial scales – the Bugs Matter citizen science survey has the potential to generate such data.

1 Introduction

A growing body of evidence (Fox et al., 2013; Hallmann et al., 2017; Goulson, D. 2019, Sánchez-Bayo et al., 2019, Thomas et al., 2019, van der Sluijs, 2020) highlights population declines in insects and other invertebrates at global scales (herein referred to collectively as 'insects'). These declines, which are evident across all functional groups of insects (herbivores, detritivores, parasitoids, predators and

pollinators) could have catastrophic impacts on the earth's natural systems and human survivability on our planet. Invertebrates are functionally of greater importance than large-bodied fauna, and in terms of biomass, bioabundance and species diversity, they make up the greatest proportion of life on earth.

Invertebrates are critical to ecosystem functions and services. They pollinate most of the world's crops, provide natural pest control services, and decompose organic matter and recycle nutrients into the soil. Without them we could not grow onions, cabbages, broccoli, chillies, most tomatoes, coffee, cocoa, most fruits, sunflowers, and rapeseed, and demand for synthetic fibres would surge because bees pollinate cotton and flax. Invertebrates underpin food chains, providing food for larger animals including birds, bats, reptiles, amphibians, fish and terrestrial mammals. Almost all birds eat insects, and many of those that eat seeds and other food as adults must feed insects to their young – it is thought to take 200,000 insects to raise a single swallow chick (Chapman et al., 2013) . Without insects, life on earth would collapse, millions of species would go extinct, and we would be surrounded by the carcases of dead animals. Consequently, considerable conservation effort is targeted at invertebrates.

Evidence of insect declines comes from targeted surveys using specific sampling techniques aimed at specific target groups. Many of these have generated long-term data sets, such as the Rothampstead Insect Survey of aphids and larger moths, since 1964 (Taylor, 1986), the UK Butterfly Monitoring Scheme, since 1976, (Brereton et al., 2020), and the National Moth Recording Scheme, since 2007 (Fox et al., 2021), and they provide a good indication of trends for those target taxa. However, generalizing regional, national and global trends from surveys of a limited number of insect groups could be inaccurate. Patterns and trends for specific species or species groups are nuanced, and while trends in some insect groups are well understood, there is a paucity of data for many others. Whilst some survey techniques such as moth trapping and butterfly transects are discriminate in terms of what species they record, there are very few established methods for large-scale monitoring of insect abundance across a broad range of insect groups. Both discriminate and indiscriminate approaches have advantages and disadvantages. Here we present the results from a survey that used an innovative method for large-scale indiscriminate monitoring of flying insect abundance , which has potential to provide an efficient, standardised and scalable approach to monitor insect population trends across local, regional and global scales.

The 'windscreen phenomenon' (Wikipedia, 2021) is a term given to the anecdotal observation that fewer insects tend to get squashed on the windscreens of cars now compared to a decade or several decades ago. This effect has been ascribed to major global declines in insect abundance. Flying insects are inadvertently sampled when they become squashed on vehicle windscreens and number plates when they are impacted. We implemented an invertebrate sampling technique based on the 'windscreen phenomenon'. The data was collected by citizen scientists to assess invertebrate abundance over a 17 year timeframe (Tinsley-Marshall et al., 2021a, 2021b). The aim was to quantify insect populations in Gwent using a standardised approach and to make comparisons with pre-existing baseline data from 2004, which was collected as part of a national survey using the same methodology led by the RSPB. By repeating the survey in Gwent in 2021 it was possible to compare the numbers of insects sampled between these points in time.

We aimed to test the null hypothesis H0: there is no evidence of variation in the numbers of insects sampled on vehicle numberplates in Gwent between 2004 and 2021 and to determine whether an alternative hypothesis H1: there is evidence of variation in the numbers of insects sampled on vehicle numberplates in Gwent between 2004 and 2021 could be accepted. This report summarises the results of an analysis of the insect abundance and participation data from the Bugs Matter survey in and around Gwent, and adds to the evidence base for patterns in invertebrate abundance locally.

2 Materials and Methods

Study area and survey design

The parameters of the study landscape were defined as the entire administrative extent of Gwent, accepting that some data would be collected from journeys that spanned the county boundary (Figure 1). It was not possible to isolate at which point in each journey insects were sampled, therefore each complete journey was included, where journeys crossed borders.

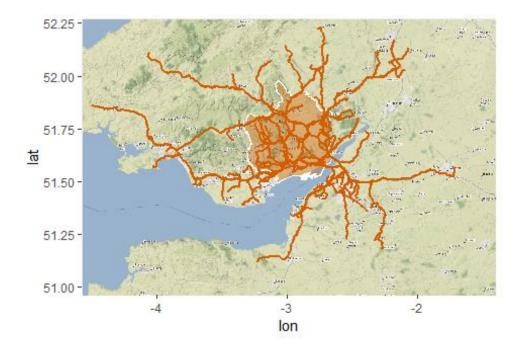


Figure 1. A map showing the distribution and extent of journeys included in this analysis of Bugs Matter survey data on insect numbers sampled by vehicle number plates for the county of Gwent

The focus of this research was defined as invertebrate abundance, acting as a proxy for the ecosystem services provided by insect populations, and as a metric for insect population change. Survey design was informed by a list of desirable attributes of monitoring programmes, ordered from most elemental to most aspirational (Pocock et al., 2015) and aimed to ensure that all relevant attributes were adopted.

Insect sampling method

Prior to commencing a journey, citizen scientists cleaned the front number plate of their vehicle to remove any residual insects. Insects were then sampled when they collided with the number plate throughout the duration of a journey. Citizen scientists were asked to participate only on essential journeys and not to make journeys specifically to take part in the survey. Using a standardised sampling grid, termed a 'splatometer', citizen scientists recorded the number of insects and other invertebrates squashed on the number plate of their car (Figure 2). Only insects within the cut-out portions of the splatometer were counted to ensure all counts were made from within a standardized area on each number plate. In 2021, data was collected on journeys undertaken between 1st June and 31st August,

and in 2004 data was collected in June. In 2004, the start and end times and locations of the journeys were recorded, along with the journey distance using vehicle odometer readings. In 2021 the precise route of the journey was recorded in real-time using the Bugs Matter mobile app.



Figure 2. Photograph showing the splatometer positioned over a number plate.

Bugs Matter mobile app

Data were submitted by citizen scientists via the Bugs Matter mobile app (Figure 3). The app provided a platform to record counts of insects on number plates, track the journey route using GPS, and collect information on the length, duration, and average speed of each journey undertaken as part of the survey. It also used an Application Programming Interface (API) number plate look-up service to collect information about vehicles involved in the survey. This data was used in the analysis to determine whether and how vehicle specifications influence insect sampling.



Figure 3. Screenshots of the Bugs Matter mobile app.

Collating explanatory variables

Time of day was calculated for each journey as the intermediate time between the start and end times. As 97.9% of journeys occurred during daytime hours (05:00-21:00), we treated time as a continuous variable in the statistical modelling, rather than converting to a factor variable or sin/cos time. The 'sf' package (Pebesma, 2018) in R was used to calculate journey length. The average speed of the journey was calculated by dividing the journey distance by the journey duration. The vehicle type, acquired via the API, was classified to align with the analysis conducted by the RSPB in 2004. These categories were car, heavy goods vehicle (HGV), multi-purpose vehicle (MPV), sports car, sports utility vehicle (SUV) and van. Data collected prior to 2021 contained only start and end postcodes, and so journey routes were obtained from the Google Directions API, through the R 'mapsapi' package (Dorman, 2022). Mean temperature was calculated for each journey by averaging the intersecting raster cell values from 0.1 degree E-OBS gridded daily mean temperature (Cornes et al., 2018). A maximum greenest pixel composite of normalized difference vegetation index (NDVI) values was generated in Google Earth Engine (Gorelick et al., 2017) from MODIS Terra Vegetation Indices 16-Day Global 250 m data (Didan, 2015) acquired between 1st January 2017 and 30th December 2021. We assumed the spatial variation in NDVI values in this composite to be representative of broad vegetation patterns across all years. NDVI describes the difference between visible and near-infrared reflectance of vegetation cover based on chlorophyll content, and can be used to estimate vegetation productivity. Artificially-surfaced areas such as roads and buildings show as low values, whilst vegetated areas show as high values. The NDVI values were averaged within a 500 m buffer of each journey route to approximate the suitability of the habitat for insects surrounding each journey route. The NDVI values were rescaled to a -10-10 range to aid interpretation of the model coefficients.

Statistical analysis

Data cleaning and preparation

To make the data comparable between journeys, insect counts were converted to a splat rate, using the ratio count/distance, expressed in a unit of splats per mile. Differences in insect splat rates (splats per mile) between years were visualized in a boxplot. In addition, relationships between other variables, such as how journey distance or the types of vehicles used in the surveys varied between years, were examined visually in boxplots and correlation plots, and tested using Kruskal-Wallis tests or Spearman correlation tests.

Prior to the analysis, some steps were taken to clean the data and remove outliers. Journeys with GPS errors were removed from the 2021 data. These errors were caused by a drop-out of background tracking due to GPS signal being lost by the device, and they appeared as long straight lines between distant locations. All journeys with a 1 km or greater gap between route vertices were omitted. Some journeys were very short with extremely high splat rates. Therefore, very short journeys of less than 0.3 miles were removed, as they are highly likely to be the result of GPS errors or incorrect use of the app, for example by the user forgetting to press the start journey button at the appropriate time. Similarly, all journeys that lasted less than one minute and journeys with an average speed of less than 1 mph or over 80 mph were omitted. In addition, all journeys during which rainfall occurred were omitted from the dataset due to the risk that rainfall could dislodge insects from numberplates, leading to bias in the data.

Modelling

A negative binomial generalised linear model (NBGLM) was used to examine the relative effects of survey year, time of day of each journey, average journey temperature, average journey speed, journey distance, vehicle type, local NDVI, and road type, on the number of insects sampled by vehicles. The response variable in our analysis was the insect count which showed a right-skewed distribution due to the high number of zero and low values, as is typical for count-derived data (Appendix 1). A NBGLM was chosen because it is appropriate for over-dispersed count data. Overdispersion was confirmed using a test for overdispersion on a Poisson model (Cameron and Trivedi, 1990), which resulted in a test statistic of c = 3.565, indicating overdispersion (c = 0 for equidispersion). The analysis was performed using the MASS package (Venables and Ripley, 2002) in RStudio (R Core Team, 2021) following established techniques (Sokal & Rolf, 1995; Crawley, 2007).

Journey distance was included in the models as an offset term. Offset terms are included in models of count-derived data to deal with counts made over different observation periods. This is preferable to using the splat rate (distance-normalizing the count using the ratio count/distance) because by adding the denominator of the ratio (distance) as an offset term, it makes use of the correct probability distributions. It can be thought of as explicitly modelling the expected rate of sampling an insect as distance driven increases. The model with offset does model the ratio (splats per mile), but in a way that is likely to be much more compatible with the data (Coelho et al., 2020).

After running the model, variance inflation factor (VIF) scores were calculated to check for multicollinearity between independent variables. A VIF score greater than 10 indicates high collinearity, which means two or more independent variables are correlated with one another. This can cause unreliable predictions and weaken the statistical power of the model. The goodness of fit of the model was assessed using McFadden's pseudo-R2 value, 1- (residual deviance)/(null deviance), which indexes the improvement of the fitted model over a model with no explanatory variables (the null model) (Hensher and Stopher, 2021). McFadden's pseudo-R2 values were compared between a model with only

survey year included as an independent variable, and the full model, to evaluate the contribution of the other independent variables to the model fit. The results of the NBGLM show the quantity of change (a multiplier) in the response variable given a one-unit change in the independent variable, while holding other variables in the model constant. These values are called incidence rate ratios and they are presented in a forest plot.

3 Results

Insect abundance

In Gwent in 2004, 1169 insects were sampled over 112 journeys comprising 5964 miles. In Gwent in 2021, 340 insects were sampled over 175 journeys comprising 2695 miles. The average splat rate in 2004 was 0.195 splats per mile and in 2021 it was 0.153 splats per mile. The spread of the insect splat rate data is shown in Figure 4. The proportion of journeys in which zero insects were sampled was 8.9% in 2004 and 34.3% in 2021. The majority of journeys (92%) were undertaken in a conventional car with the remainder being undertaken in HGV, MPV, Van (Appendix 2). The average time of day of journeys in 2004 was 14:41 whilst in 2021 it was 13:16 (Appendix 3). The mean average journey speed in 2004 was 39.1 mph, whilst in 2021 it was 27.5 mph (Appendix 4). The average journey temperature in 2004 was 16.1°C, whilst in 2021 it was 16.4°C (Appendix 5). The average journey distance in 2004 was 53.2 miles and in 2021 the average journey distance was 15.4 miles (Appendix 6). The average NDVI surrounding journeys in 2004 was 5.848 and in 2021 it was 7.233 (Appendix 7). A positive correlation was observed between journey distance and count of splats (Appendix 8). A positive correlation was also observed between journey distance and splat rate (splats per mile) (Appendix 9). The VIF scores showed very low collinearity between independent variables.

The results of the NBGLM show that in and around Gwent there were 39.8% (95% CI [12.8%, 58.5%]) fewer insects sampled on vehicle number plates in 2021 than in 2004, and this difference was statistically significant (p = 0.0093982) (Figure 5). McFadden's pseudo-R2 value was 0.048, and in a model with only year as a predictor it was 0.021. This shows that the amount of deviance accounted for increased from 2.1% to 4.8% by including the other variables.

Regarding the other explanatory variables, the results showed no significant relationship between splat rate and the type of vehicle, time of day, mean temperature, journey distance, local NDVI or average speed of the journey.

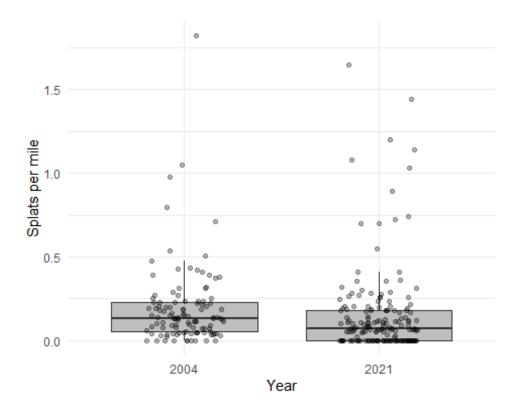


Figure 4. Box and whisker plot with jittered data points showing the spread of insect splat rate data (splats per mile) from the Bugs Matter survey of insects on car number plates in Gwent in each of the survey years. Red points are high-leverage observations that were omitted from the NBGLM. The boxes indicate the interquartile range (central 50% of the data), either side of the median splat rate which is shown by the horizontal line inside the box. The vertical lines extend out by 1.5 times the interquartile range, and the data points themselves are added with a 'horizontal jitter' so they do not overlap to improve visualization of the data distribution. If splat rate on every journey was identical, we would only see the line across the middle of the box, with the data points on top of it.

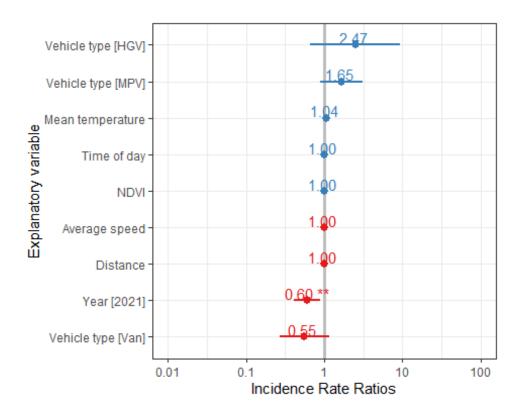


Figure 5. Forest plot of incidence rate ratios from the NBGLM of Bugs Matter survey data of insects on car number plates in Gwent, showing the quantity of change (a multiplier) in splat rate (splats per mile) given a one-unit change in the independent variable, while holding other variables in the model constant. Significant relationships between splat rate and independent variables are shown by asterisks (* p < 0.05, ** p < 0.01, *** p < 0.001). Vehicle types are compared to the reference category of 'conventional cars'. The reference year is 2004.

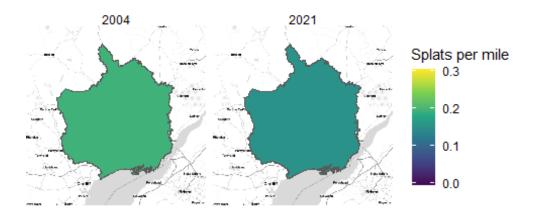


Figure 6. Heat map of splat rate from the Bugs Matter survey of insects on car number plates in Gwent in each of the survey years, 2004 - 2021.

Participation

A total of 63 citizen scientists signed-up to the Bugs Matter survey in Gwent in 2021, which was substantially fewer than the other three participating counties. The majority signed-up in the initial launch period between mid-May and early-June, although there were considerable spikes in signups around key dates. For example, the slight increase in early June coincides with Bugs Matter featuring on BBC Springwatch. There was a slight lag between launch and sign-up spikes due to delays in translating communication materials into Welsh. Unlike the other counties, Gwent received no new signups after early August (Figure 7).

Of the 63 individuals who signed up to the Bugs Matter app, 20 participated in the survey, the criteria for which was submitting data for at least one journey. We can calculate a conversion rate, as the number of participants that submitted one or more journeys (20) divided by the number of sign-ups (63). This gives a conversion rate of 32%. In comparison to the other participating Trusts, Gwent had the lowest number of signups (63) but the second-lowest number of journeys (175) submitted via the app. The average number of journeys recorded by each surveyor was 8.8. This means that, on average, each surveyor in Gwent undertook more journeys than elsewhere in the UK. The number of journeys submitted via the app rose steadily throughout the survey season and did not drop-off into August (Figure 8).

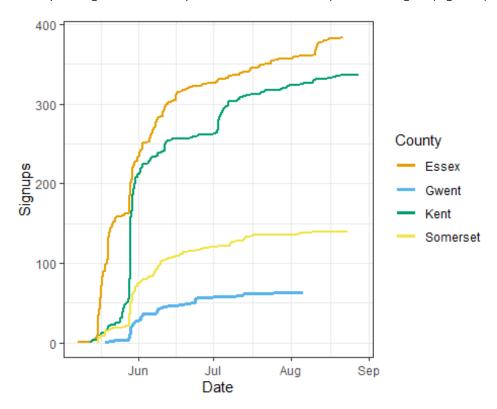


Figure 7. Number of signups to the Bugs Matter app in Gwent compared to other participating counties during the 2021 survey season.

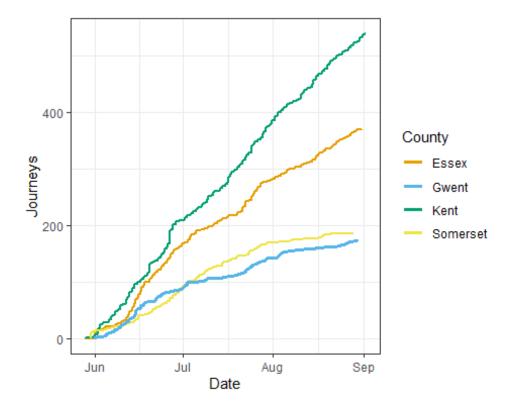


Figure 8. Number of journeys submitted via the Bugs Matter app in Gwent compared to other participating counties during the 2021 survey season.

4 Discussion

Insect abundance

The results of this study suggest a pattern of reduction in numbers of insects sampled on vehicle numberplates in Gwent, consistent with those reported elsewhere by others (Fox et al., 2013; Goulson, D., 2019; Hallmann et al., 2017). The estimate of change in splat rate between 2004 and 2021 (a decrease of 39.8%) has a lower confidence interval of 12.8% and an upper confidence interval of 58.5%, at a 95% confidence level. This means that if we repeated the study, 95% of the time we would expect the estimate of change in splat rate to fall between these values. However, it should be noted that the observations reported here are based on data from only three points in time and with a skewed temporal distribution, and consequently do not constitute a trend and we do not interpret this as a decline. With such a low temporal resolution, there is a risk of uncharacteristically high or low insect abundances during these sampling years showing a trend that is unrepresentative of actual insect abundance patterns. To accurately estimate change in insect abundance over time, the population needs to be monitored comprehensively at regular intervals over an extended timeframe to reveal the direction and scale of genuine trends. However, the pattern observed in this study is consistent with examples of insect decline reported elsewhere and and informs a growing requirement for conservation research, policy and practice targeted at invertebrates in the UK. Similar declines were noted in a study that sampled insects splatted on vehicle windscreens between 1997 and 2017 in the Netherlands (Møller, 2019).

Insect population dynamics are influenced by a range of natural factors that vary inter-annually and across various spatial and temporal scales (Figure 9). These factors add noise to longer-term trends in

insect abundance but can be partly controlled for in our modelling. For instance, the inclusion of mean temperature and NDVI in our models controls for inter-annual differences in temperature and spatial variation in vegetation cover, both of which may naturally influence insect abundance. Whilst insect populations vary spatially and temporally, so did our insect sampling approach. The time of day and date of the journey, the vehicle type, the vehicle speed and the journey distance all create sampling bias, which we have attempted to control for in our methods, by measuring these variables and including them in our models (Figure 9). By controlling for these effects we obtain more accurate estimates of change in insect splat rate between survey years. However, there are other important variables that are not yet included in the models. For example, environmental variables with demonstrated lethal and sub-lethal influence on insect population ecology such as pesticide use, pollution, land-use change and climate change could explain a further proportion of the unexplained variation in the data. Our model also lacks data on a number of other influential factors such as variation in habitat type and management, disease and predation of insects, other weather conditions including humidity or wind, and natural variation in insect lifecycles or flight periods. Finally, there may be subtle differences in survey methods and/or approaches between journeys and/or years which were not recorded or communicated to subsequent survey managers.

By including a range of variables in the statistical model, it was possible to examine how specific variables affected insect splat rate while controlling for the effects of the other variables in the model. This was important for a more robust estimation of change in splat rate between years, but also allowed us to examine the effects of other factors on insect splat rate. Whilst the amount of deviance explained by the model increased with the addition of other variables (2.1% to 4.8%), no significant relationship was found between splat rate and the type of vehicle, the time of day, mean temperature, journey distance, local NDVI, or average speed of the journey.

Splat rate showed no correlation with journey distance, as shown by a significant but non-directional result. This is somewhat expected as the splat rate is normalized over journey distance, however we might have expected to see more insects sampled over longer journeys due to the increased chances of encountering areas with higher densities of flying insects. Conversely, longer journeys tend to follow motorways where insect abundance may be lower and it is possible that sampled insects could be blown off the number plate on long journeys, especially if the average journey speed is high. The correlation plots showing the relationship between journey distance and splat count and rate (Appendix 8) show some long journeys with very few insect splats or low overall splat rates, which could be partially explained by this phenomenon.

Insect sampling was restricted to transects along the road network, and therefore the spatial coverage of the surveys is inherently limited. Whilst this design serves to provide a robust measure of change in insect abundance, by comparing one year to the next, we caution against the use of this data for estimating true insect abundance. Another limitation of the survey was that citizen scientists may have forgotten to clean their numberplate prior to conducting a survey, although the risk of this is very low for the 2021 Bugs Mater survey, where the app required a checkbox confirmation that the number plate had been cleaned. We are currently investigating how we could use AI algorithms to automatically count the number of insects on number plates. This would use a virtual template in the app, similar to those used to automatically read credit card details, and return the count in real-time to the user. This would negate the requirement for a splatometer making it easier for citizen scientists to count and record data.

Participation

Sign-ups in Gwent were lower than the other participating Wildlife Trusts, but the reasons for this are not fully known. One possibility, is that the delay in communicating the launch of the survey in Welsh may have meant that national and local press were out of sync. No sign-ups occurred after mid-August, whereas for other counties this was not the case, and so it may be worthwhile reviewing the communication strategy for August.

The number of journeys submitted via the app occurred at a fairly constant rate over the survey season, suggesting that participants remained motivated throughout the summer. Moreover, the number of journeys recorded by each participant was high, with an average of 9 journeys per surveyor. However, a large number of participants in Gwent signed up but did not complete a single journey (43/63), hence a low conversion rate of 32%. To determine the cause of this, a post-survey questionnaire was circulated to participants, which showed that the delay in receiving the splatometer was a key reason. Consequently, priorities for the app going forward are to include automatic application of the splatometer through the smartphone, as well as other improvements to the communications and implementation strategy for the 2022 survey.

Synthesis and Application

The Bugs Matter survey successfully quantified a difference in the number of insects sampled on vehicle numberplates over time from baseline data established in 2004. The approach has the potential to provide an efficient, standardised and scalable approach to monitor insect population trends across local, regional and global scales, to add to the growing body of evidence for trends in insect populations and to provide a coarse metric of the functional provision by insects within ecosystems.

An increasing number of studies are accumulating evidence of insect declines, and associated consequence for ecosystem functions. It is important to recognise that these patterns and trends are often nuanced, and that local conditions and choice of analytical approach may mean that results reported locally or regionally may not reflect patterns everywhere. The media uptake of negative trends from short time series data such as those presented here for example, may be acting to exaggerate the perception of an 'insect Armageddon', with potential consequences for public confidence in research. We recognise and stress that the results we have reported here do not constitute a trend, and advocate strongly for data collection over extended timeframes to enable conclusions about trends in insect populations to be drawn. We believe that the widespread adoption of the Bugs Matter survey facilitated by the Bugs Matter app can provide a replicable and scalable approach for the generation of an enhanced evidence-base that can be used to assess trends and drive positive action for insects and other invertebrates.

Increasing sample size both by increasing the number of citizen scientist participants and the number of journeys undertaken would provide greater confidence in the reliability of our data as a robust indicator of patterns in insect abundance. Similarly, cross-validating our results with other monitoring schemes for insect abundance, such as the Rothamsted Insect Survey (RIS) (Fox et al., 2013) or the results of long-term Malaise trapping studies (Hallmann et al., 2017), would provide another means to calibrate and critique the patterns in our data. There is potential for the survey method to have global application and relevance, and deployed at a national scale, it can provide data at resolutions appropriate to the scale at which the ecosystem services provided by insects operate. By continuing to promote participation in the survey in subsequent years, insect conservationists can capitalise on the opportunity to gather long-term data and build the evidence base for insect abundance at UK county and national scale.

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Author contributions

This project was led and managed by PTM, who contributed much of the text for this report along with guidance for the statistical analyses. LB led on the collation of variables, statistical analysis and report writing, whilst RS performed initial data cleaning and formatting and the analysis of the participation data. AR and AS were part of the project team in 2019, AR leading on GIS and data assimilation, and AR on citizen scientist participation.

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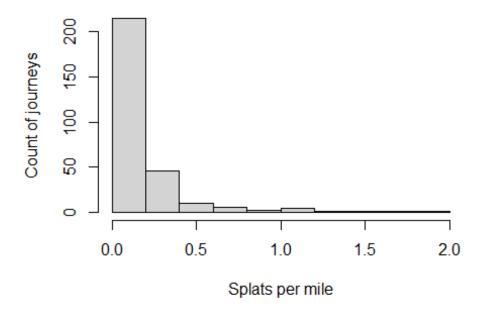
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Appendices

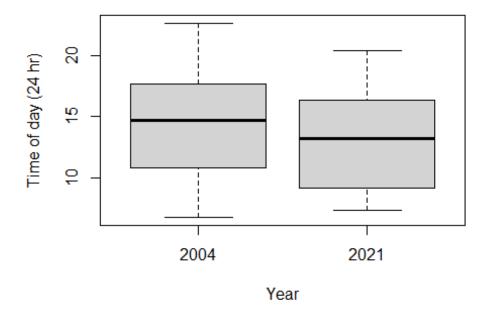
Appendix 1. A histogram of the splat rate (splats per mile) data.



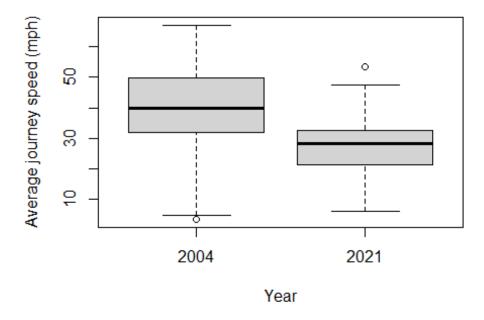
Appendix 2. The number of journeys conducted by each vehicle type in each survey year.

	2004	2019	2021
Car	101	0	164
HGV	1	0	1
MPV	1	0	10
Sports car	0	0	0
SUV	0	0	0
Van	9	0	0

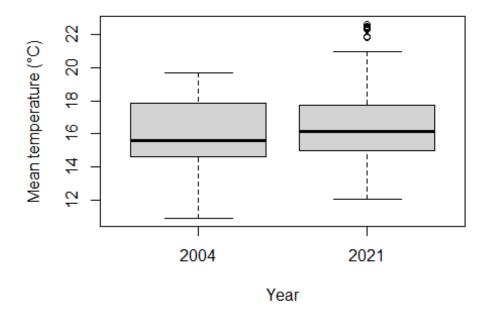
Appendix 3. Box and whisker plot showing the spread of the time of day of journey data from the Bugs Matter survey of insects on car number plates in Gwent in each of the survey years. A Kruskal-Wallis test showed a significant difference in the time of day at which journeys were undertaken between the survey years (H(1) = 6.926, p = 0.0084945).



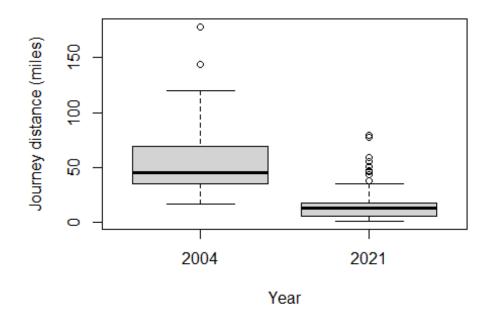
Appendix 4. Box and whisker plot showing the spread of the average journey speed data from the Bugs Matter survey of insects on car number plates in Gwent in each of the survey years. A Kruskal-Wallis test showed a significant difference in the average journey speed between the survey years (H(1) = 60.825, p = < 0.001).



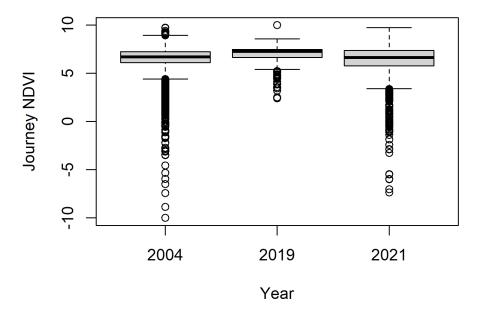
Appendix 5. Box and whisker plot showing the spread of the mean journey temperature data from the Bugs Matter survey of insects on car number plates in Gwent in each of the survey years. A Kruskal-Wallis test showed no significant difference in the mean journey temperature between the survey years (H(1) = 0.424, p = 0.51504).



Appendix 6. Box and whisker plot showing the spread of the journey distance data from the Bugs Matter survey of insects on car number plates in Gwent in each of the survey years. A Kruskal-Wallis test showed a significant difference in the journey distances between the survey years (H(1) = 160.182, p = < 0.001).

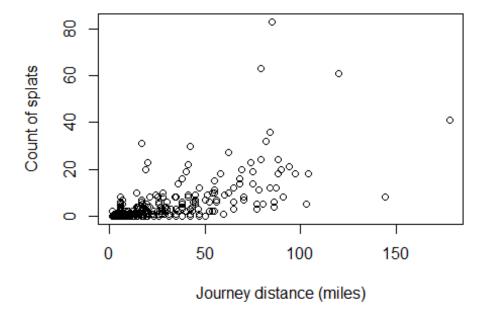


Appendix 7. Box and whisker plot showing the spread of the NDVI data from the Bugs Matter survey of insects on car number plates in Gwent in each of the survey years. A Kruskal-Wallis test showed a significant difference in the journey NDVI between the survey years (H(1) = 144.134, p = < 0.001).



Appendix 8. Correlation plot showing the relationship between journey distance (x-axis) and count of splats (y-axis). A Spearman correlation test showed a significant positive correlation between journey

distance and count of splats (rho = 0.666, p = < 0.001).



Appendix 9. Correlation plot showing the relationship between journey distance (x-axis) and splat rate (y-axis). A Spearman correlation test showed a weak but significant positive correlation between journey distance and count of splats (rho = 0.211, p = < 0.001).

